Our group has recently developed novel nano-sized drug carriers that spatially target a tumour and release their payload in the presence of ultrasound-induced inertial cavitation. To maximize drug release and distribution within the tumour, co-localisation of the drug carrier and cavitation nuclei is necessary. We have recently demonstrated that rough-patterned silica nanoparticles can reduce inertial cavitation thresholds to clinically relevant levels, and will extravasate in tumours alongside the liposomes by virtue of their size. We now report on the underlying mechanisms that these nanoparticles, which are orders of magnitude smaller than the acoustic wavelength, can instigate inertial cavitation. The rough surface of the nanoparticle is modelled as a plane with a crevasse that traps a nanobubble. Using this model, we predict the motion of a gas bubble as it emerges from the cavity in response to the compressional and rarefactional ultrasonic pressures. We show that cavitation occurs when the nanobubble breaks free from the surface, growing unstably before collapsing during the compressional half cycle of the acoustic wave. Calculations show that a nanoscaled cavity greatly reduces the cavitation threshold across all frequencies and geometries studied. In addition, cavitation thresholds non-linearly decrease with increasing cavity size.
INTRODUCTION

Cancer is one of the leading causes of death in the western world (Jemal et al., 2011). Recently, there have been significant advancements in the efficacy of chemotherapeutic drugs. However, there is still an inherent problem with delivering such drugs to the entirety of the tumour. This problem is typically overcome by increasing dosages that result in unpleasant and potentially lethal side effects (Helmy, 2006). Furthermore, tumours can recur (Bartelink et al., 2001), becoming aggressive and metastatic (Engel et al., 2003) if not completely removed or treated.

In order to address such issues, researchers are developing tumour-targeting techniques that take advantage of the unique physiology of the tumour and thereby lower the required dosages (Kratz and Warnecke, 2012). Tumours typically grow quickly and this is accompanied by rapid proliferation of new blood vessels. Angiogenesis results in an irregular vascular structure, which is inherently “leaky”, with nano-sized gaps between endothelial cells. Additionally, the intratumoural space is pressurised, which prevents small particles from diffusing into the tumour. This combination of leaky vasculature and intratumoural pressure results in a self-selection of particles, typically between 50 and 400 nm in diameter that can enter a tumour. This phenomenon is known as the enhanced permeability and retention effect (EPR) (Iyer et al., 2006).

Typically, the EPR effect prevents most small drugs and larger drug carriers from entering deep into the tumour. However, our group has recently developed a unique nano-sized drug carrier that will naturally target tumours due to the EPR effect and release its payload when disrupted by ultrasound-induced inertial cavitation. To maximize the efficacy of this drug delivery method, it is necessary to co-localise both the drug carrier and the cavitation nuclei. In order to provide co-localisation, the cavitation agent must be on the nano-scale, but up until now, the only known cavitation agents are on the micron scale (Sirsi, 2009). Recently, our research group has demonstrated that rough-patterned silica nanoparticles will reduce the thresholds required for inertial cavitation (Wagstaffe et al., 2012). In this manuscript we report on the fundamental mechanisms that allow such nano-particles the capacity to initiate inertial cavitation. In short, the patterning on the surface of the nanoparticles trap nanobubbles, which will expand and contract with the negative and positive pressure cycles of the acoustic wave. With sufficient force, we propose here that these nanobubbles can grow orders of magnitude larger than the surface cavity, detach, and subsequently cavitate inertially. In the present work, we investigate the effect of ultrasound frequency and of the characteristic dimension of surface roughness on the pressure threshold required to instigate inertial cavitation from such nanoparticles.

THEORY

Surfaces with micro- and nano-scopic crevasses will entrap bubbles, preventing their otherwise inevitable dissolution into the surrounding medium. Our rough-patterned nanoparticles trap nanobubbles that respond to pressure perturbations due to acoustic waves. We model the trapped nanobubble, with a radius of curvature, R, by assuming that the crevasse lies in an infinite plane and has an arbitrary wall geometry, w, as shown in figure (1a) (Chappell and Payne, 2007).

![Figure 1](image.png)

**FIGURE 1.** Geometry of a nanobubble within a crevasse (a) and emerging from a crevasse (b).
During the rarefactual period of the acoustic wave, it is possible that the nanobubble escapes the crevasse and grows beyond the surface, as shown in figure (1b). If either the momentum of the bubble growth, or the size of the bubble generates a contact angle, $\theta$, approaching the receding contact angle, then the nanobubble will detach from the surface. Once free, the nanobubble will either immediately dissolve into the surrounding medium, or, inertially collapse due to the compressional forces of the ultrasound wave.

In order to predict the behaviour of the nanobubble in response to ultrasound, we utilise a modified Raleigh-Plesset model (equation 1).

$$ R \dddot{R} + \frac{3}{2} R \ddot{R} = \frac{1}{\rho_L} \left( P_L(R) - P^c(t) - P_H \right). $$  

(1)

where, $R = f(x)$, $x$ is the height of the three phase contact line, $\dot{R}$ and $\ddot{R}$ are the bubble wall radial velocity and acceleration respectively, $\rho_L$ is the liquid density, $P_L$ is the gas-liquid interfacial pressure, $P^c$ is the acoustic pressure, $t$ is time, and $P_H$ is the hydrostatic pressure. Equation 1 was solved using an ordinary differential equation solver in Matlab. In all of the simulations, the acoustic pulse was modelled as a five-cycle sine wave. Additionally, we are only considering the three phase contact angle as the parameter that determines nanobubble detachment. The crevasse in all simulations presented here is a cylinder ($w(x) = r$, where $r$ is the cavity radius) with depth of $2r$. The initial position of the nanobubble within the cavity was arbitrarily chosen to be 90% of the total depth of the cavity.

RESULTS

Utilising the theory described above, we have simulated a single nanobubble trapped in a cylindrical crevasse over a variety of acoustic pressures, frequencies, and crevice sizes. Throughout the simulations, the nanobubble interfacial radial velocity was calculated and used as the metric for inertial cavitation; collapse occurred when bubble wall velocity was greater than the speed of sound. Thus, we predicted the necessary conditions required for inertial cavitation by simulating nanobubble behaviour across a variety of parameters. Here we demonstrate the effects of acoustic pressure, frequency, and cavity size. In figure 2, we show that the required acoustic pressure for inertial cavitation nonlinearly decreases as the cavity size increases. However, these results also suggest that cavities in the size range 10-50 nm will readily cavitate if driven by pressure amplitudes of a few MPa for any frequency in the range 0.5-3.3 MHz. Figure 2 also shows the response to changes in frequency, indicating that as the frequency is increased, the required pressures for inertial cavitation increase. The effects of frequency, however, are only pronounced at larger crevasse sizes.

![Figure 2](image-url)

**FIGURE 2.** The threshold for inertial cavitation for various cavity sizes and ultrasound frequencies (0.5, 1, 1.7, and 3.3 MHz) are shown.

Figure 3 shows representative responses of a nanobubble driven at a frequency of 3.3 MHz and with pressure amplitudes, which either do not or do yield inertial collapse. For non-cavitation conditions (figure 3a), the nanobubble will oscillate either solely within the cavity, or move above and below the surface. In the cavitation condition regime (figure 3b), the nanobubble will detach from the walls or surface of the crevice. Once detached, there is a sudden violent expansion of the nanobubble corresponding to volumetric increases of several orders of magnitude.
magnitude. Collapse occurs during the compressional cycle of the acoustic wave coupled to the momentum of the surrounding fluid.

**FIGURE 3.** The response of a nanobubble trapped in a 30 nm radius cylindrical crevasse with a 60 nm depth to a 5 cycle 3.3 MHz ultrasound pulse at (a) 1 MPa and (b) 3 MPa. (a) At 1 MPa, the nanobubble does not detach from the cavity, or show any signs of inertial cavitation. Instead, the nanobubble oscillates from inside to outside the cavity. (b) At 3 MPa, the nanobubble immediately breaks free from the cavity. The nanobubble next grows unstably and collapses during the compressional phase of the ultrasound wave.

**DISCUSSION**

One of the greatest barriers to comprehensive chemotherapy is the ability to non-invasively treat the entirety of a tumour without damaging systemic side effects. Our research group has recently developed a nano sized drug delivery vehicle that will naturally accumulate in tumours and release its payload exclusively in the presence of inertial cavitation. Unfortunately, inertial cavitation is chaotic and spatially unpredictable. Cavitation agents such as microbubbles are too large to localise with the drug delivery vehicle or to extravasate through the endothelial gaps into the intratumoural space. We have recently demonstrated that nano-scopic rough patterned silica may instigate inertial cavitation, and by virtue of their size will remain with the nano-drug carrier. Our simulations indicate that these silica nanoparticles contain entrapped gases that, when exposed to ultrasound, will detach from the cavity, expand several orders of magnitude, and inertially collapse during the positive pressure half-cycle. This massive expansion indicates that it is plausible to observe and therefore confirm this hypothesis. We also demonstrate that it is possible that nanocavities will reduce the pressure thresholds for inertial cavitation to clinically relevant levels, which corresponds to the experimental evidence from our lab. As a result, these silica nanoparticles may be a novel tool for instigating inertial cavitation in a clinical environment, and warrants further investigation.

In our current simulations, several assumptions have been made as first order approximations. For example, the volumetric expansions are assumed to occur radially and axi-symmetrically. In reality, the nanobubble moves unilaterally away from the base of the crevasse. As a result, these simulations provide an underestimate of nanobubble volumetric growth and compression, indicating that nucleation of inertial cavitation may require even lower pressure thresholds than currently predicted. Similarly, we are neglecting the momentum of the nanobubble as it emerges from the cavity as a detachment criterion, which will also further lower thresholds for inertial cavitation. Finally, we have assumed that the bubble movement within the cavity was sufficiently fast such that the influence of the static thermodynamics of the gas-liquid-solid interface (i.e., advancing and receding contact angles) on nanobubble movement is neglected. As a result, the influence of contact angle only affects the detachment of the nanobubble. The effects of such assumptions are currently unknown and offer another avenue of exploration.

**CONCLUSION**

In this manuscript we report on the development of underlying fundamental mechanisms that govern the behaviour of rough patterned silica nanoparticles when perturbed by ultrasound. By modelling the rough pattern on the surface of the nanoparticle as a crevice of cylindrical geometry, we have shown that nanobubble detachment from the crevice is a necessary condition for inertial cavitation. Additionally, the acoustic pressures required for inertial cavitation decrease nonlinearly with increasing cavity size. These preliminary results give evidence that rough patterned silica nanoparticles will yield inertial cavitation for pressure amplitudes of a few MPa in the low MHz range and can thus provide a suitable release mechanism for our novel nanoparticulate drug carriers.
REFERENCES


